

Figure 1A Nucleotide sequence of inserted environmental DNA (*mHKcel* cellulase)

ATCTTTAGAG	ATGAGCGTGA	TAGTCAGTGG	ACCTTAAAAG	AACTTAAAAG	50
ATTTTTAGAA	GAGATTAAAA	CGGACCCGCA	CCATCTCTCT	GTTTATTTTG	100
ATGGGGGATT	TGATTTGGAG	ACACAACGAT	CTGGCCTTGG	GTGTGTGATT	150
TATTATGAAC	AAAATGACAC	GTCTTACCGG	GTGAGAAAAA	ACGCTACCGT	200
GGCGTCATTG	ACATCGAATA	ACCAAGCAGA	ATATGCCGCT	TTGCATTTAG	250
GACTTAAAAG	AACTTGAAGG	GATCGGGCGC	ATCATCTGCC	TATCACCATT	300
TACGGTGATT	CTCAAGTTGT	GATCAATCAG	TAAAAGGAG	AATGGGCCGT	350
GTATGGAGGA	GGTGTAAAT	AAATGGGCTG	ACCCGTATTG	ATCAAGCATT	400
TAGCTAAATT	AGGCATGACC	GCTACTTATA	AGTTAATCCC	CCGTAAAGAA	450
AACCGCGAAG	CCGATCAACT	GGCTACACAA	GCGTTAAACG	GGCAAGAAAT	500
TATAAGTCAA	CGTGATATCA	GTGAGCGTGG	TGCAGATTAG	GCTGCACCGC	550
GCAAAAAAAG	TCAACGTGTT	TAGGAATGGA	CAGGGATTAA	AGCAACATAA	600
TTCTCTCTAA	GCAAACGTTG	CGACAGCAAG	AGAGAAGCAT	ATAAGGTTTT	650
TCTGAGTTAG	TCTATTTATA	CCAATGTCCA	CGTACTAAAT	AAACCTCTCA	700
TCAAAGTGGA	TTTTTTTGATT	AATTCAC TTC	CACTCCTACC	TTTATCTATA	750
TAAATTAGTT	CCTTTTTTGT	TAATAATCAC	TAATTTTGCC	GGTATTTTTT	800
AATAGAAATA	TATGCTAGAT	TATAAACTAG	TAGCCGTATA	GAAGGTGGTG	850
ATTGCCCTTA	TAAGAGACGT	CTGGCAAACA	TAAAAGCATC	GCATTATTAT	900
AATCGAAAGG	TGGAGATGAG	ACATGGGTTA	TACCCAAGCT	AAGTGTATGG	950
TGAAAAAAAC	GGTCTTGTTT	GGTTTAATTC	TCTGTTTAGG	TGTGTCAATG	1000
TTTGTACCAG	TTACATCAGC	TGAAGATAGG	GTCTCTTCGT	CACAGGTGGA	1050
TATCCAATCA	TATGTAGCAG	ATATGCAACC	TGGCTGGAAT	TTAGGTAATA	1100
CATTTGATGC	GATAGGAGAT	GATGAAACAG	CATGGGGAAA	CCCTCGTGTA	1150
ACGAGAGAAT	TAATAGAAAT	GATTGCTGAT	GAAGGGTATA	AAAGTATTTCG	1200
TATCCCAGTC	ACATGGCAAA	ATCAAATGGG	TGGTTCTCCA	GATTATACAA	1250
TTAATGAAGA	TTATATCAAG	CGGGTAGAGC	AAGTGATAGA	TTGGGCGTTG	1300
GAGGAAGACT	TGTATGTGAT	GTTAAATGTG	CATCATGACT	CATGGCTGTG	1350
GATGTATGAT	ATGGAACATA	ACTATGATGA	GGTGATGGCA	AGATATACAG	1400
CTATTTGGGA	ACAATTGTCG	GAAAAATTCA	AAAACCACTC	CCATAAGTTG	1450
ATGTTTGAGA	GTGTCAATGA	GCCTAGGTTT	ACGCAGGAGT	GGGGAGAGAT	1500
TCAAGAAAAT	CATCATGCTT	ACTTAGAAGA	TTTAAATAAG	ACGTTCTATT	1550
ATATTGTCAG	AGAGTCAGGA	GGCAATAATG	TGGAGCGCCC	TTAGTATTG	1600
CCTACGATAG	AAACAGCCAC	GTCTCAGGAT	TTACTAGATC	GCTTGTATCA	1650
AACAATGGAA	GACTTGATG	ACCCTCATTT	AATTGCCACG	GTCATTATT	1700
ATGGCTTTTG	GCCCTTTAGT	GTCAATATAG	CAGGGTACAC	CCGTTTTGAA	1750
CAGGAGACAC	AACAAGATAT	TATAGACACG	TTTGACCGTG	TTCATAACAC	1800
ATTTACAGCG	AATGGGATCC	CAGTTGTATT	AGGTGAATTT	GGTTTGTTAG	1850
GCTTTGATAA	AAGTACGGAC	GTCATTCAGC	AAGGTGAGAA	ATTAAAATTT	1900
TTTGAGTTTC	TCATCCATCA	TCTCAATGAA	CGTGATATAA	CCCATATGTT	1950
ATGGGATAAC	GGTCAGCATT	TAAAGCGAGA	AACTTATTCA	TGGTATGATC	2000
AGGAATTTCA	TGACATATTA	AAAGCGAGTT	GGGAGGGGCG	TTCTGCTACA	2050
GCTGAGTCTA	ATTTTCATTCA	TGTGAAGGAC	GGAGAGCCAA	TTAGAGATCA	2100
ACATATACAG	CTTTACTTAA	ACGGAAATGA	GCTAACTGCC	CTACAGGCAG	2150
GGGACGAATC	GCTTGTACTA	GGAGAGGATT	ATGAGCTAGC	AGGAGACGTA	2200
TTAACGCTAA	AAGCGGGCAT	CCTCACAAGA	TTAATTACCC	CTGGCCAATT	2250
AGGAACGAAT	GCGGTCATCA	CAGCTCAATT	TAATTCTGGA	GCAGACTGGC	2300
GTTTTCAATT	ACAGAATGTG	GACGTGCCAA	CAGTCGAAAA	TACAGATGGC	2350
TCAATATGGC	ATTTTGCGAT	CCCTACCCAT	TTAATGGTG	ATAGTCTTGC	2400

Figure 1B

GACGATGGAA	GCTGTTTATG	CAAACGGAGA	ATATGCTGGC	CCGCAAGATT	2450
GGACGTCATT	TAAAGAATTT	GGCGAGGCGT	TTTCCCCTAA	TTACGCCACA	2500
GGGGAAATTA	TTATAACAGA	AGCCTTCTTT	AACGCGGTAC	GGGATGATGA	2550
TATCCATTTA	ACATTTTCATT	ATTGGAGCGG	AGAGACGGTG	GAATATACAT	2600
TACGTAAAAA	TGGAAATTAT	GTTCAAGGTA	GACGGTAACA	TGATTTTAAAT	2650
TAATAGATAA	AACAGCCTAC	CTATCGTTTT	TGGAAGAAGG	CAAACGAATC	2700
TCATCTTACC	AACACCGTGC	TTTAGAACTT	TAGAAGTGAC	GGTGTTTTTT	2750
AAGACATGAG	GAGAGACAAT	CCTCTATCAA	CAGTCACCAA	TTTTTATTCA	2800
GGAGGTGTCA	AGTTATCTAA	CGTTCTATGA	ATGCATATAG	TTTCTGACGA	2850
ATAAACATAG	TTAAAAAGAA	GTGAGCCTAG	TTCCCGAGGG	GAAGGGGATA	2900
ATGCCAACGT	ATTGGATTAA	AGTACCTTCT	TGATAAAAAG	AAAGGGTTTT	2950
CAAGAGGTGG	AAATGGGCTC	GTTTGTTATA	CTTTAATTAC	ACCTTGGAAC	3000
GTCATTTTGG	CGGTGCTACT	TAGTAAGATG	ACTGACATCA	TAAAAGAGGA	3050
GTGGGTTCGA	TGGCTTTAAT	TCAATTAAGC	TTTAAATCAC	GAGCATTAAAT	3100
GTTGCAAACC	TCTGTCAATG	TTTTATTACC	GGTGGGAATG	AATGCGGTAG	3150
ATTTTACACC	AAGTGATGAT	TTTTCTTATG	TTACTGACCC	TTTTCCTGTC	3200
CTATATCTTT	TGCATGGTGC	AACTGATGAT	TATTTCAGCAT	GGCTACGTCT	3250
GTCCTCCATT	GAACGATATG	CTGAAGAAAA	AAAATTGGCG	GTCGTCATGC	3300
CAAATGCTGA	TATGAGTGCG	TATACGGATA	TGGTACATGG	ACATCGTTAC	3350
TGGACGTATA	TTAGTAAGGA	GCTGCCTGAG	TTTATCAAAG	CGACTTTTCC	3400
TATTTCTCAG	CACCGTGAAG	ACACCTTTGC	GGCTGGTCTG	TCTATGGGAG	3450
GATACGGGGC	TTTTAAATTA	GCGTTGCGGC	AACCGGAACG	CTTCGCTGCA	3500
GNTGTGTCAT	TATCAGGTGC	AGTTGATATG	AGAGAAGCAA	GTCAACCAGA	3550
CTCCCTATTT	GTGAACGCAT	TTGGTGAAGG	GACGAAAATC	GCAGGGACAG	3600
ATCTTGATCT	TTTTCATTTA	ATTAAAAAGT	TGGGGGTATA	TGAAGGGGCT	3650
AAACCAGCCC	TTTTTCAAGC	GTGTGGGACA	GAGGACTTTT	TATATGAAGA	3700
TAATGTGAGA	TTTAGAGATT	ATGCACGACA	AGTGAATGCC	GATTTAACTT	3750
ATGAAGAAGG	TCCTGGTGGT	CATGAATGGG	CTTATTGGGA	TAGAAT	3796

Figure 2 ORF Nucleotide sequence of mHKcel cellulase gene

ATGGGTTATA	CCCAAGCTAA	GTGTATGGTG	AAAAAACCGG	TCTTGTTTGG	50
TTTAATTCCTC	TGTTTAGGTG	TGTCAATGTT	TGTACCAGTT	ACATCAGCTG	100
AAGATAGGGT	CTCTTCGTCA	CAGGTGGATA	TCCAATCATA	TGTAGCAGAT	150
ATGCAACCTG	GCTGGAATTT	AGGTAATACA	TTTGATGCGA	TAGGAGATGA	200
TGAAACAGCA	TGGGGAAACC	CTCGTGTAAC	GAGAGAATTA	ATAGAAATGA	250
TTGCTGATGA	AGGGTATAAA	AGTATTTCGT	TCCCAGTCAC	ATGGCAAAAT	300
CAAAATGGGTG	GTTCTCCAGA	TTATACAATT	AATGAAGATT	ATATCAAGCG	350
GGTAGAGCAA	GTGATAGATT	GGGCGTTGGA	GGAAGACTTG	TATGTGATGT	400
TAAATGTGCA	TCATGACTCA	TGGCTGTGGA	TGTATGATAT	GGAACATAAC	450
TATGATGAGG	TGATGGCAAG	ATATACAGCT	ATTTGGGAAC	AATTGTCGGA	500
AAAATTCAAA	AACCACTCCC	ATAAGTTGAT	GTTTGAGAGT	GTCAATGAGC	550
CTAGGTTTAC	GCAGGAGTGG	GGAGAGATT	AAGAAAATCA	TCATGCTTAC	600
TTAGAAGATT	TAAATAAGAC	GTTCTATTAT	ATTGTCAGAG	AGTCAGGAGG	650
CAATAATGTG	GAGCGCCCTT	TAGTATTGCC	TACGATAGAA	ACAGCCACGT	700
CTCAGGATTT	ACTAGATCGC	TTGTATCAAA	CAATGGAAGA	CTTGATGAC	750
CCTCATTTAA	TTGCCACGGT	TCATTATTAT	GGCTTTTGGC	CCTTTAGTGT	800
CAATATAGCA	GGGTACACCC	GTTTTGAACA	GGAGACACAA	CAAGATATTA	850
TAGACACGTT	TGACCGTGTT	CATAACACAT	TTACAGCGAA	TGGGATCCCA	900
GTTGTATTAG	GTGAATTTGG	TTTGTTAGGC	TTTGATAAAA	GTACGGACGT	950
CATTCAGCAA	GGTGAGAAAT	TAAAAATTTT	TGAGTTTCTC	ATCCATCATC	1000
TCAATGAACG	TGATATAACC	CATATGTTAT	GGGATAACGG	TCAGCATTTA	1050
AAGCGAGAAA	CTTATTCATG	GTATGATCAG	GAATTTTCATG	ACATATTAAA	1100
AGCGAGTTGG	GAGGGGCGTT	CTGCTACAGC	TGAGTCTAAT	TTCATTCATG	1150
TGAAGGACGG	AGAGCCAATT	AGAGATCAAC	ATATACAGCT	TTACTTAAAC	1200
GGAAATGAGC	TAAGTGCCCT	ACAGGCAGGG	GACGAATCGC	TTGTACTAGG	1250
AGAGGATTAT	GAGCTAGCAG	GAGACGTATT	AACGCTAAAA	GCGGGCATCC	1300
TCACAAGATT	AATTACCCCT	GGCCAATTAG	GAACGAATGC	GGTCATCACA	1350
GCTCAATTTA	ATTCTGGAGC	AGACTGGCGT	TTTCAATTAC	AGAATGTGGA	1400
CGTGCCAACA	GTCGAAAATA	CAGATGGCTC	AATATGGCAT	TTTGCGATCC	1450
CTACCCATTT	TAATGGTGAT	AGTCTTGCGA	CGATGGAAGC	TGTTTATGCA	1500
AACGGAGAAT	ATGCTGGCCC	GCAAGATTGG	ACGTCATTTA	AAGAATTTGG	1550
CGAGGCGTTT	TCCCCTAATT	ACGCCACAGG	GGAAATTATT	ATAACAGAAG	1600
CCTTCTTTAA	CGCGGTACGG	GATGATGATA	TCCATTTAAC	ATTTTATTAT	1650
TGGAGCGGAG	AGACGGTGGA	ATATACATTA	CGTAAAAATG	GAAATTATGT	1700
TCAAGGTAGA	CGGTAA				1715

Figure 3 Amino acid sequence of cellulase mHKcel

MGYTQAKCMV KKTVLFGIL CLGVSMFVPV TSAEDRVSSS QVDIQSYVAD	50
MQPGWNLGNT FDAIGDDETA WGNPRVTREL IEMIADEGYK SIRIPVTWQN	100
QMGGSPTYI NEDYIKRVEQ VIDWALEEDL YVMLNVHHDS WLWMYDMEHN	150
YDEVMARYTA IWEQLSEKFK NHSHKLMFES VNEPRFTQEW GEIQENHHAY	200
LEDLNKTFYY IVRESGGNNV ERPLVLPTIE TATSQDLLDR LYQTMEDLDD	250
PHLIATVHYY GFWPFSVNIA GYTRFEQETQ QDIIDTFDRV HNTFTANGIP	300
VVLGEFGLLG FDKSTDVIQQ GEKLKFFEFL IHHLNERDIT HMLWDNGQHL	350
KRETYSWYDQ EFHDILKASW EGRSATAESN FIHVKDGEPI RDQHIQLYLN	400
GNELTALQAG DESLVLGEDY ELAGDVLTLK AGILTRLITP GQLGTNAVIT	450
AQFNSGADWR FQLQNVDVPT VENTDGSIIH FAIPTHFNGD SLATMEAVYA	500
NGEYAGPQDW TSFKEFGCAF SPNYATGEII ITEAFFNAVR DDDIHLTFHY	550
WSETVEYTL RKNNGNYVQGR R	571

Figure 4 . Enzyme Activity with Increasing NaCl Concentration

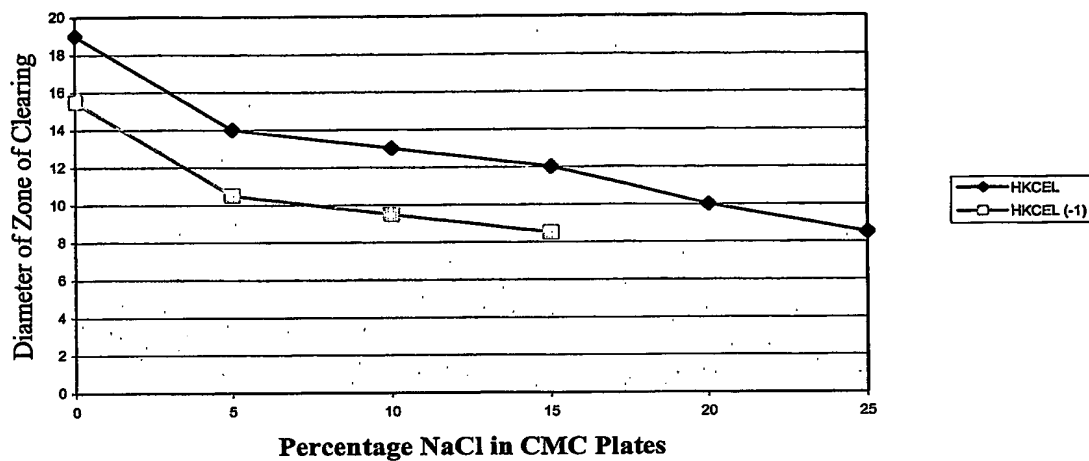


Figure 5. Influence of pH on mHKcel Cellulase Activity

